

## **RAW SEQUENCE LISTING**

**The Biotechnology Systems Branch of the Scientific and Technical  
Information Center (STIC) no errors detected.**

Application Serial Number: 10/658, 355C  
Source: IFW/b  
Date Processed by STIC: 11/15/2006

# ***ENTERED***



IFW16

## RAW SEQUENCE LISTING

DATE: 11/15/2006

PATENT APPLICATION: US/10/658,355C

TIME: 11:44:02

Input Set : F:\923SEQ.004.txt

Output Set: N:\CRF4\11152006\J658355C.raw

3 <110> APPLICANT: Gantier, Rene  
 4 Guyon, Thierry  
 5 Hugo, Cruz Ramos  
 6 Vega, Manuel  
 7 Drittanti, Lila  
 9 <120> TITLE OF INVENTION: Rational Directed Protein Evolution Using Two Dimensional  
 10 Rational Mutagenesis Scanning  
 12 <130> FILE REFERENCE: 17019-013001/923  
 14 <140> CURRENT APPLICATION NUMBER: US/10/658,355C  
 15 <141> CURRENT FILING DATE: 2003-09-08  
 17 <150> PRIOR APPLICATION NUMBER: 60/457,063  
 18 <151> PRIOR FILING DATE: 2003-03-21  
 20 <150> PRIOR APPLICATION NUMBER: 60/410,258  
 21 <151> PRIOR FILING DATE: 2002-09-09  
 23 <160> NUMBER OF SEQ ID NOS: 502  
 24 <170> SOFTWARE: FastSEQ for Windows Version 4.0  
 26 <210> SEQ ID NO: 1  
 27 <211> LENGTH: 165  
 28 <212> TYPE: PRT  
 29 <213> ORGANISM: Homo sapiens  
 31 <400> SEQUENCE: 1  
 32 Cys Asp Leu Pro Gln Thr His Ser Leu Gly Ser Arg Arg Thr Leu Met  
 33 1 5 10 15  
 35 Leu Leu Ala Gln Met Arg Arg Ile Ser Leu Phe Ser Cys Leu Lys Asp  
 36 20 25 30  
 38 Arg His Asp Phe Gly Phe Pro Gln Glu Glu Phe Gly Asn Gln Phe Gln  
 39 35 40 45  
 41 Lys Ala Glu Thr Ile Pro Val Leu His Glu Met Ile Gln Gln Ile Phe  
 42 50 55 60  
 44 Asn Leu Phe Ser Thr Lys Asp Ser Ser Ala Ala Trp Asp Glu Thr Leu  
 45 65 70 75 80  
 47 Leu Asp Lys Phe Tyr Thr Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu  
 48 85 90 95  
 50 Ala Cys Val Ile Gln Gly Val Gly Val Thr Glu Thr Pro Leu Met Lys  
 51 100 105 110  
 53 Glu Asp Ser Ile Leu Ala Val Arg Lys Tyr Phe Gln Arg Ile Thr Leu  
 54 115 120 125  
 56 Tyr Leu Lys Glu Lys Lys Tyr Ser Pro Cys Ala Trp Glu Val Val Arg  
 57 130 135 140  
 59 Ala Glu Ile Met Arg Ser Phe Ser Leu Ser Thr Asn Leu Gln Glu Ser  
 60 145 150 155 160  
 61 Leu Arg Ser Lys Glu  
 62 165

CP2-67

## RAW SEQUENCE LISTING

DATE: 11/15/2006

PATENT APPLICATION: US/10/658,355C

TIME: 11:44:02

Input Set : F:\923SEQ.004.txt

Output Set: N:\CRF4\11152006\J658355C.raw

```

64 <210> SEQ ID NO: 2
65 <211> LENGTH: 165
66 <212> TYPE: PRT
67 <213> ORGANISM: Artificial Sequence
69 <220> FEATURE:
70 <223> OTHER INFORMATION: D2A Mutant INF-alpha 2b
72 <400> SEQUENCE: 2
73 Cys Ala Leu Pro Gln Thr His Ser Leu Gly Ser Arg Arg Thr Leu Met
74 1 5 10 15
76 Leu Leu Ala Gln Met Arg Arg Ile Ser Leu Phe Ser Cys Leu Lys Asp
77 20 25 30
79 Arg His Asp Phe Gly Phe Pro Gln Glu Glu Phe Gly Asn Gln Phe Gln
80 35 40 45
82 Lys Ala Glu Thr Ile Pro Val Leu His Glu Met Ile Gln Gln Ile Phe
83 50 55 60
85 Asn Leu Phe Ser Thr Lys Asp Ser Ser Ala Ala Trp Asp Glu Thr Leu
86 65 70 75 80
88 Leu Asp Lys Phe Tyr Thr Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu
89 85 90 95
91 Ala Cys Val Ile Gln Gly Val Gly Val Thr Glu Thr Pro Leu Met Lys
92 100 105 110
94 Glu Asp Ser Ile Leu Ala Val Arg Lys Tyr Phe Gln Arg Ile Thr Leu
95 115 120 125
97 Tyr Leu Lys Glu Lys Lys Tyr Ser Pro Cys Ala Trp Glu Val Val Arg
98 130 135 140
100 Ala Glu Ile Met Arg Ser Phe Ser Leu Ser Thr Asn Leu Gln Glu Ser
101 145 150 155 160
102 Leu Arg Ser Lys Glu
103 165
105 <210> SEQ ID NO: 3
106 <211> LENGTH: 165
107 <212> TYPE: PRT
108 <213> ORGANISM: Artificial Sequence
110 <220> FEATURE:
111 <223> OTHER INFORMATION: P4A Mutant INF-alpha 2b
113 <400> SEQUENCE: 3
114 Cys Asp Leu Ala Gln Thr His Ser Leu Gly Ser Arg Arg Thr Leu Met
115 1 5 10 15
117 Leu Leu Ala Gln Met Arg Arg Ile Ser Leu Phe Ser Cys Leu Lys Asp
118 20 25 30
120 Arg His Asp Phe Gly Phe Pro Gln Glu Glu Phe Gly Asn Gln Phe Gln
121 35 40 45
123 Lys Ala Glu Thr Ile Pro Val Leu His Glu Met Ile Gln Gln Ile Phe
124 50 55 60
126 Asn Leu Phe Ser Thr Lys Asp Ser Ser Ala Ala Trp Asp Glu Thr Leu
127 65 70 75 80
129 Leu Asp Lys Phe Tyr Thr Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu
130 85 90 95
132 Ala Cys Val Ile Gln Gly Val Gly Val Thr Glu Thr Pro Leu Met Lys

```

## RAW SEQUENCE LISTING

DATE: 11/15/2006

PATENT APPLICATION: US/10/658,355C

TIME: 11:44:02

Input Set : F:\923SEQ.004.txt

Output Set: N:\CRF4\11152006\J658355C.raw

```

133          100          105          110
135 Glu Asp Ser Ile Leu Ala Val Arg Lys Tyr Phe Gln Arg Ile Thr Leu
136          115          120          125
138 Tyr Leu Lys Glu Lys Lys Tyr Ser Pro Cys Ala Trp Glu Val Val Arg
139          130          135          140
141 Ala Glu Ile Met Arg Ser Phe Ser Leu Ser Thr Asn Leu Gln Glu Ser
142 145          150          155          160
143 Leu Arg Ser Lys Glu
144          165
146 <210> SEQ ID NO: 4
147 <211> LENGTH: 165
148 <212> TYPE: PRT
149 <213> ORGANISM: Artificial Sequence
151 <220> FEATURE:
152 <223> OTHER INFORMATION: Q5A Mutant INF-alpha 2b
154 <400> SEQUENCE: 4
155 Cys Asp Leu Pro Ala Thr His Ser Leu Gly Ser Arg Arg Thr Leu Met
156 1          5          10          15
158 Leu Leu Ala Gln Met Arg Arg Ile Ser Leu Phe Ser Cys Leu Lys Asp
159          20          25          30
161 Arg His Asp Phe Gly Phe Pro Gln Glu Glu Phe Gly Asn Gln Phe Gln
162          35          40          45
164 Lys Ala Glu Thr Ile Pro Val Leu His Glu Met Ile Gln Gln Ile Phe
165          50          55          60
167 Asn Leu Phe Ser Thr Lys Asp Ser Ser Ala Ala Trp Asp Glu Thr Leu
168 65          70          75          80
170 Leu Asp Lys Phe Tyr Thr Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu
171          85          90          95
173 Ala Cys Val Ile Gln Gly Val Gly Val Thr Glu Thr Pro Leu Met Lys
174          100          105          110
176 Glu Asp Ser Ile Leu Ala Val Arg Lys Tyr Phe Gln Arg Ile Thr Leu
177          115          120          125
179 Tyr Leu Lys Glu Lys Lys Tyr Ser Pro Cys Ala Trp Glu Val Val Arg
180          130          135          140
182 Ala Glu Ile Met Arg Ser Phe Ser Leu Ser Thr Asn Leu Gln Glu Ser
183 145          150          155          160
184 Leu Arg Ser Lys Glu
185          165
187 <210> SEQ ID NO: 5
188 <211> LENGTH: 165
189 <212> TYPE: PRT
190 <213> ORGANISM: Artificial Sequence
192 <220> FEATURE:
193 <223> OTHER INFORMATION: T6A Mutant INF-alpha 2b
195 <400> SEQUENCE: 5
196 Cys Asp Leu Pro Gln Ala His Ser Leu Gly Ser Arg Arg Thr Leu Met
197 1          5          10          15
199 Leu Leu Ala Gln Met Arg Arg Ile Ser Leu Phe Ser Cys Leu Lys Asp
200          20          25          30

```

## RAW SEQUENCE LISTING

DATE: 11/15/2006

PATENT APPLICATION: US/10/658,355C

TIME: 11:44:02

Input Set : F:\923SEQ.004.txt

Output Set: N:\CRF4\11152006\J658355C.raw

```

202 Arg His Asp Phe Gly Phe Pro Gln Glu Glu Phe Gly Asn Gln Phe Gln
203      35              40              45
205 Lys Ala Glu Thr Ile Pro Val Leu His Glu Met Ile Gln Gln Ile Phe
206      50              55              60
208 Asn Leu Phe Ser Thr Lys Asp Ser Ser Ala Ala Trp Asp Glu Thr Leu
209 65              70              75              80
211 Leu Asp Lys Phe Tyr Thr Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu
212      85              90              95
214 Ala Cys Val Ile Gln Gly Val Gly Val Thr Glu Thr Pro Leu Met Lys
215      100             105             110
217 Glu Asp Ser Ile Leu Ala Val Arg Lys Tyr Phe Gln Arg Ile Thr Leu
218      115             120             125
220 Tyr Leu Lys Glu Lys Lys Tyr Ser Pro Cys Ala Trp Glu Val Val Arg
221      130             135             140
223 Ala Glu Ile Met Arg Ser Phe Ser Leu Ser Thr Asn Leu Gln Glu Ser
224 145             150             155             160
225 Leu Arg Ser Lys Glu
226      165
228 <210> SEQ ID NO: 6
229 <211> LENGTH: 165
230 <212> TYPE: PRT
231 <213> ORGANISM: Artificial Sequence
233 <220> FEATURE:
234 <223> OTHER INFORMATION: H7A Mutant INF-alpha 2b
236 <400> SEQUENCE: 6
237 Cys Asp Leu Pro Gln Thr Ala Ser Leu Gly Ser Arg Arg Thr Leu Met
238 1      5              10              15
240 Leu Leu Ala Gln Met Arg Arg Ile Ser Leu Phe Ser Cys Leu Lys Asp
241      20              25              30
243 Arg His Asp Phe Gly Phe Pro Gln Glu Glu Phe Gly Asn Gln Phe Gln
244      35              40              45
246 Lys Ala Glu Thr Ile Pro Val Leu His Glu Met Ile Gln Gln Ile Phe
247      50              55              60
249 Asn Leu Phe Ser Thr Lys Asp Ser Ser Ala Ala Trp Asp Glu Thr Leu
250 65              70              75              80
252 Leu Asp Lys Phe Tyr Thr Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu
253      85              90              95
255 Ala Cys Val Ile Gln Gly Val Gly Val Thr Glu Thr Pro Leu Met Lys
256      100             105             110
258 Glu Asp Ser Ile Leu Ala Val Arg Lys Tyr Phe Gln Arg Ile Thr Leu
259      115             120             125
261 Tyr Leu Lys Glu Lys Lys Tyr Ser Pro Cys Ala Trp Glu Val Val Arg
262      130             135             140
264 Ala Glu Ile Met Arg Ser Phe Ser Leu Ser Thr Asn Leu Gln Glu Ser
265 145             150             155             160
266 Leu Arg Ser Lys Glu
267      165
269 <210> SEQ ID NO: 7
270 <211> LENGTH: 165

```

## RAW SEQUENCE LISTING

DATE: 11/15/2006

PATENT APPLICATION: US/10/658,355C

TIME: 11:44:02

Input Set : F:\923SEQ.004.txt

Output Set: N:\CRF4\11152006\J658355C.raw

```

271 <212> TYPE: PRT
272 <213> ORGANISM: Artificial Sequence
274 <220> FEATURE:
275 <223> OTHER INFORMATION: S8A Mutant INF-alpha 2b
277 <400> SEQUENCE: 7
278 Cys Asp Leu Pro Gln Thr His Ala Leu Gly Ser Arg Arg Thr Leu Met
279 1 5 10 15
281 Leu Leu Ala Gln Met Arg Arg Ile Ser Leu Phe Ser Cys Leu Lys Asp
282 20 25 30
284 Arg His Asp Phe Gly Phe Pro Gln Glu Glu Phe Gly Asn Gln Phe Gln
285 35 40 45
287 Lys Ala Glu Thr Ile Pro Val Leu His Glu Met Ile Gln Gln Ile Phe
288 50 55 60
290 Asn Leu Phe Ser Thr Lys Asp Ser Ser Ala Ala Trp Asp Glu Thr Leu
291 65 70 75 80
293 Leu Asp Lys Phe Tyr Thr Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu
294 85 90 95
296 Ala Cys Val Ile Gln Gly Val Gly Val Thr Glu Thr Pro Leu Met Lys
297 100 105 110
299 Glu Asp Ser Ile Leu Ala Val Arg Lys Tyr Phe Gln Arg Ile Thr Leu
300 115 120 125
302 Tyr Leu Lys Glu Lys Lys Tyr Ser Pro Cys Ala Trp Glu Val Val Arg
303 130 135 140
305 Ala Glu Ile Met Arg Ser Phe Ser Leu Ser Thr Asn Leu Gln Glu Ser
306 145 150 155 160
307 Leu Arg Ser Lys Glu
308 165
310 <210> SEQ ID NO: 8
311 <211> LENGTH: 165
312 <212> TYPE: PRT
313 <213> ORGANISM: Artificial Sequence
315 <220> FEATURE:
316 <223> OTHER INFORMATION: L9A Mutant INF-alpha 2b
318 <400> SEQUENCE: 8
319 Cys Asp Leu Pro Gln Thr His Ser Ala Gly Ser Arg Arg Thr Leu Met
320 1 5 10 15
322 Leu Leu Ala Gln Met Arg Arg Ile Ser Leu Phe Ser Cys Leu Lys Asp
323 20 25 30
325 Arg His Asp Phe Gly Phe Pro Gln Glu Glu Phe Gly Asn Gln Phe Gln
326 35 40 45
328 Lys Ala Glu Thr Ile Pro Val Leu His Glu Met Ile Gln Gln Ile Phe
329 50 55 60
331 Asn Leu Phe Ser Thr Lys Asp Ser Ser Ala Ala Trp Asp Glu Thr Leu
332 65 70 75 80
334 Leu Asp Lys Phe Tyr Thr Glu Leu Tyr Gln Gln Leu Asn Asp Leu Glu
335 85 90 95
337 Ala Cys Val Ile Gln Gly Val Gly Val Thr Glu Thr Pro Leu Met Lys
338 100 105 110
340 Glu Asp Ser Ile Leu Ala Val Arg Lys Tyr Phe Gln Arg Ile Thr Leu

```

RAW SEQUENCE LISTING ERROR SUMMARY  
PATENT APPLICATION: US/10/658,355C

DATE: 11/15/2006  
TIME: 11:44:03

Input Set : F:\923SEQ.004.txt  
Output Set: N:\CRF4\11152006\J658355C.raw

Please Note:

Use of n and/or Xaa have been detected in the Sequence Listing. Please review the Sequence Listing to ensure that a corresponding explanation is presented in the <220> to <223> fields of each sequence which presents at least one n or Xaa.

Seq#:195; N Pos. 1,22

**VERIFICATION SUMMARY**

PATENT APPLICATION: US/10/658,355C

DATE: 11/15/2006

TIME: 11:44:03

Input Set : F:\923SEQ.004.txt

Output Set: N:\CRF4\11152006\J658355C.raw

L:7601 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:195 after pos.:0